

CLAIMS

We claim:

1. An isolated polypeptide of at least 15 amino acid residues comprising an epitope-bearing portion of a protein of SEQ ID NO:2.

2. The polypeptide of claim 1 wherein said polypeptide comprises a segment that is at least 70% identical to a sequence selected from the group consisting of:

(a) residues 52-179 of SEQ ID NO:2; and

(b) residues 258-370 of SEQ ID NO:2.

3. The isolated polypeptide according to claim 1 wherein said polypeptide is selected from the group consisting of:

residues 19-179 of SEQ ID NO:2;

residues 52-179 of SEQ ID NO:2;

residues 19-253 of SEQ ID NO:2;

residues 52-253 of SEQ ID NO:2;

residues 19-255 of SEQ ID NO:2;

residues 52-255 of SEQ ID NO:2;

residues 19-257 of SEQ ID NO:2;

residues 52-257 of SEQ ID NO:2;

residues 19-253 of SEQ ID NO:2;

residues 52-253 of SEQ ID NO:2;

residues 19-370 of SEQ ID NO:2;

residues 52-370 of SEQ ID NO:2;

residues 180-370 of SEQ ID NO:2; and

residues 258-370 of SEQ ID NO:2.

4. An isolated polypeptide comprising a sequence of amino acids of the formula $R1_x-R2_y-R3_z$, wherein:

R1 is a polypeptide of from 100 to 130 residues in length, is at least 70% identical to residues 52-179 of SEQ ID NO:2, and comprises a sequence motif C[KR]Y[DNE][WYF]X{11,15}G[KR][WYF]C (SEQ ID NO:4) corresponding to residues 109-131 of SEQ ID NO:2;

R2 is a polypeptide at least 90% identical to residues 180-257 of SEQ ID NO:2;

R3 is a polypeptide at least 70% identical in amino acid sequence to residues 258-370 of SEQ ID NO:2 and comprises cysteine residues at positions corresponding to residues 272, 302, 306, 318, 360, and 362 of SEQ ID NO:2; a glycine residue at a position corresponding to residue 304 of SEQ ID NO:2; and a sequence motif CX{18,33}CXGXCX{6,33}CX{20,50}CXC (SEQ ID NO:3) corresponding to residues 272-362 of SEQ ID NO:2; and

each of x, y, and z is individually 0 or 1, subject to the limitations that:
at least one of x and z is 1; and
if x and z are each 1, then y is 1.

5. The isolated polypeptide of claim 4 wherein x=1.
6. The isolated polypeptide of claim 5 wherein R1 is at least 90% identical to residues 52-179 of SEQ ID NO:2.
7. The isolated polypeptide of claim 5 wherein R1 comprises residues 52-179 of SEQ ID NO:2.
8. The isolated polypeptide of claim 5 wherein R1 is at least 90% identical to residues 19-179 of SEQ ID NO:2.
9. The isolated polypeptide of claim 5 wherein y=1.
10. The isolated polypeptide of claim 9 wherein z=1.
11. The isolated polypeptide of claim 10 wherein R3 is at least 90% identical to residues 258-370 of SEQ ID NO:2.
12. The isolated polypeptide of claim 4 wherein said polypeptide comprises residues 52-253 of SEQ ID NO:2, residues 180-370 of SEQ ID NO:2, or residues 52-370 of SEQ ID NO:2.
13. The isolated polypeptide of claim 4 wherein z=1.
14. The isolated polypeptide of claim 13 wherein R3 is at least 90% identical to residues 258-370 of SEQ ID NO:2.

15. The isolated polypeptide of claim 13 wherein R3 comprises residues 258-370 of SEQ ID NO:2.

16. The isolated polypeptide of claim 13 wherein $y=1$.

17. The isolated polypeptide of claim 16 wherein $x=1$ and R1 is at least 90% identical to residues 52-179 of SEQ ID NO:2.

18. The isolated polypeptide of claim 17 wherein R3 is at least 90% identical to residues 258-370 of SEQ ID NO:2.

19. The isolated polypeptide of claim 13, further comprising cysteine residues at positions corresponding to residues 308 and 316 of SEQ ID NO:2.

20. The isolated polypeptide of claim 4, further comprising an affinity tag.

21. The isolated polypeptide of claim 4, further comprising an immunoglobulin constant domain.

22. An isolated protein comprising a first polypeptide operably linked to a second polypeptide, wherein said first polypeptide comprises a sequence of amino acids of the formula $R1_x-R2_y-R3_z$, wherein:

R1 is a polypeptide of from 100 to 130 residues in length, is at least 70% identical to residues 52-179 of SEQ ID NO:2, and comprises a sequence motif $C[KR]Y[DNE][WYF]X\{11,15\}G[KR][WYF]C$ (SEQ ID NO:4) corresponding to residues 109-131 of SEQ ID NO:2;

R2 is a polypeptide at least 90% identical to residues 180-257 of SEQ ID NO:2;

R3 is a polypeptide at least 70% identical in amino acid sequence to residues 258-370 of SEQ ID NO:2 and comprises cysteine residues at positions corresponding to residues 272, 302, 306, 318, 360, and 362 of SEQ ID NO:2; a glycine residue at a position corresponding to residue 304 of SEQ ID NO:2; and a sequence motif $CX\{25,33\}CXGXCX\{10,33\}CX\{20,50\}CXC$ (SEQ ID NO:3) corresponding to residues 272-362 of SEQ ID NO:2; and

each of x , y , and z is individually 0 or 1, subject to the limitations that:

at least one of x and z is 1; and

if x and z are each 1, then y is 1,

and wherein said protein modulates cell proliferation, apoptosis, differentiation, metabolism, or migration.

23. The isolated protein of claim 22 wherein said protein is a heterodimer.

24. The isolated protein of claim 23 wherein z is 1 and said second polypeptide is selected from the group consisting of VEGF, VEGF-B, VEGF-C, VEGF-D, zveg3, PlGF, PDGF-A, and PDGF-B.

25. The isolated protein of claim 23 wherein:

x=1, z=1, and said second polypeptide comprises residues 46-345 of SEQ ID

NO:32;

x=1 and said second polypeptide comprises residues 46-170 of SEQ ID

NO:32; or

z=1 and said second polypeptide comprises residues 235-345 of SEQ ID

NO:32.

26. The isolated protein of claim 22 wherein said protein is a homodimer.

27. The isolated protein of claim 26 wherein z=1.

28. The isolated protein according to claim 27 wherein each of said first and second polypeptides comprises residues 258-370 of SEQ ID NO:2.

29. The isolated protein of claim 26 wherein x=1.

30. The isolated protein of claim 29 wherein each of said first and second polypeptides comprises residues 52-179 of SEQ ID NO:2.

31. An isolated protein produced by a method comprising:

(a) culturing a host cell containing an expression vector comprising the following operably linked elements:

a transcription promoter;

a DNA segment encoding a polypeptide comprising a sequence of amino acid residues selected from the group consisting of:

i) residues 52-370 of SEQ ID NO:2;

ii) residues 52-253 of SEQ ID NO:2;

iii) residues 180-370 of SEQ ID NO:2; and

iii) residues 258-370 of SEQ ID NO:2; and

a transcription terminator,

under conditions whereby the DNA segment is expressed; and

(b) recovering from the cell the protein product of expression of the DNA construct.

32. An isolated polynucleotide of up to approximately 4.4 kb in length, wherein said polynucleotide encodes a polypeptide comprising a sequence of amino acids of the formula $R1_x-R2_y-R3_z$, wherein:

R1 is a polypeptide of from 100 to 130 residues in length, is at least 70% identical to residues 52-179 of SEQ ID NO:2, and comprises a sequence motif $C[KR]Y[DNE][WYF]X\{11,15\}G[KR][WYF]C$ (SEQ ID NO:4) corresponding to residues 109-131 of SEQ ID NO:2;

R2 is a polypeptide at least 90% identical to residues 180-257 of SEQ ID NO:2;

R3 is a polypeptide at least 70% identical in amino acid sequence to residues 258-370 of SEQ ID NO:2 and comprises cysteine residues at positions corresponding to residues 272, 302, 306, 318, 360, and 362 of SEQ ID NO:2; a glycine residue at a position corresponding to residue 304 of SEQ ID NO:2; and a sequence motif $CX\{25,33\}CXGX\{10,33\}CX\{20,50\}CXC$ (SEQ ID NO:3) corresponding to residues 272-362 of SEQ ID NO:2; and

each of x, y, and z is individually 0 or 1, subject to the limitations that:

at least one of x and z is 1; and

if x and z are each 1, then y is 1.

33. The polynucleotide of claim 32, wherein said polynucleotide is DNA.

34. The polynucleotide of claim 33 comprising nucleotides 1 through 1110 of SEQ ID NO:6.

35. The polynucleotide of claim 33 comprising nucleotides 226 through 1335 of SEQ ID NO:1.

36. An expression vector comprising the following operably linked elements:

a transcription promoter;

a DNA polynucleotide according to claim 32, and
a transcription terminator.

37. The expression vector of claim 36, further comprising a secretory signal sequence operably linked to the DNA polynucleotide.

38. A cultured cell into which has been introduced an expression vector according to claim 36, wherein said cell expresses the polypeptide encoded by the DNA polynucleotide.

39. A pharmaceutical composition comprising a protein according to claim 22 in combination with a pharmaceutically acceptable vehicle.

40. The pharmaceutical composition of claim 39 wherein $x=1$.

41. The pharmaceutical composition of claim 39 wherein $z=1$.

42. A method of producing a protein comprising:
culturing a cell into which has been introduced an expression vector according to claim 36, whereby said cell expresses the polypeptide encoded by the DNA segment; and
recovering the expressed protein.

43. An antibody that specifically binds to an epitope of a polypeptide according to claim 4.

44. The antibody of claim 43 which is a monoclonal antibody.

45. The antibody of claim 43 which is a single-chain antibody.

46. The antibody of claim 43 operably linked to a reporter molecule.

47. A method for detecting a genetic abnormality in a patient, comprising:
obtaining a genetic sample from a patient;
incubating the genetic sample with a polynucleotide comprising at least 14 contiguous nucleotides of SEQ ID NO:1 or the complement of SEQ ID NO:1, under conditions wherein said polynucleotide will hybridize to a complementary polynucleotide sequence, to produce a first reaction product; and

comparing said first reaction product to a control reaction product, wherein a difference between said first reaction product and said control reaction product is indicative of a genetic abnormality in the patient.

48. A polypeptide comprising a sequence selected from the group consisting of:

residues 46-234 of SEQ ID NO:33 operably linked to residues 258-370 of SEQ ID NO:2;

residues 46-170 of SEQ ID NO:33 operably linked to residues 180-370 of SEQ ID NO:2;

residues 52-257 of SEQ ID NO:2 operably linked to residues 235-345 of SEQ ID NO:33; and

residues 52-179 of SEQ ID NO:2 operably linked to residues 171-345 of SEQ ID NO:33.

49. A method of activating a cell-surface PDGF receptor, comprising exposing a cell comprising a cell-surface PDGF receptor to the polypeptide or protein of any of claims 1-31, whereby the polypeptide or protein binds to and activates the receptor.

50. The method of claim 49 wherein the receptor is a PDGF alpha-receptor.

51. The method of claim 49 wherein the receptor is a PDGF beta-receptor.

52. A method of inhibiting a PDGF receptor mediated cellular process, comprising exposing a cell comprising a cell-surface PDGF receptor to a compound that inhibits binding of the polypeptide or protein of any of claims 1-31 to the receptor.

53. A method of stimulating the growth of bone tissue, comprising applying to bone a growth-stimulating amount of the polypeptide or protein of any of claims 1-31.

54. A method of modulating the proliferation, differentiation, migration, or metabolism of bone cells, comprising exposing bone cells to an effective amount of the polypeptide or protein of any of claims 1-31.